

Serial No.: 09/829,251
Filed: April 9, 2001

REMARKS

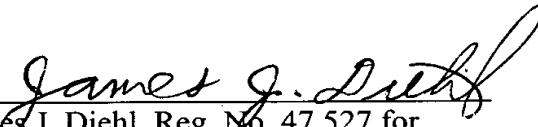
The specification has been amended to delete reference to figures in the parent application that were inadvertently omitted upon filing the present application. The description of the omitted figures has been moved to the experimental section of the specification and re-written to avoid specific reference to the figures or to information that could not be obtained without the figures. Applicants submit that these amendments introduce no new matter.

Applicants submit that the application is now in form for examination and subsequent allowance. If the Examiner believes there are issues remaining that may be disposed of by telephone, he/she is invited to call the undersigned attorney at (415) 781-1989.

Respectfully submitted,

FLEHR HOHBACH TEST
ALBRITTON & HERBERT LLP

Dated: Nov. 30, 2001


James J. Diehl, Reg. No. 47,527 for
Richard F. Trecartin, Reg. No. 31,801

Four Embarcadero Center
Suite 3400
San Francisco, CA 94111-4187
Telephone: (415) 781-1989

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VERSION SHOWING CHANGES MADE

The text from page 4, line 33 to page 5, line 22 (paragraphs beginning at page 4, line 33, page 5, line 3 and page 5, line 11) has been cancelled. The paragraph beginning at page 19, line 33, was amended as follows:

Each 0.5 OD₆₀₀ pellet was then prepared for gel analysis as follows. Each pellet was resuspended in 50 μ l TE (10mM Tris pH7.6, 1mM EDTA). After the addition of 10 μ l 10% SDS, 5 μ l reducing agent (1M dithiothreitol or 1M β -mercaptoethanol), the samples were heated at about 90°C for 2 minutes and then vortexed. Samples were allowed to cool to room temperature, after which 500 μ l acetone was added. The samples were vortexed and then left at room temperature for about 15 minutes. Samples were centrifuged for 5 minutes. The supernatants were discarded, and the pellets resuspended in 20 μ l water, 5 μ l reducing agent, 25 μ l NOVEX 2X sample buffer. Samples were heated at about 90°C for 3-5 minutes, then vortexed. After centrifugation for 5 minutes, supernatants were transferred to clean tubes and the pellets discarded. 5-10 μ l of each sample was loaded onto 10 well, 1.0 mm NOVEX manufactured gel (San Diego, CA.) and electrophoresed for 1.5-2 hr at 120 volts. Gels were stained with Coomassie blue to visualize polypeptide (Figures 19-21).--

The following three paragraphs were inserted preceding the paragraph beginning at page 20, line 11 of the specification as originally filed:

-- In one case, a polypeptide gel was run and stained with Coomassie blue to show secretion of mature ICAM-1 in E. coli under control of variant STII signal sequences. A TIR of relative strength 9 was provided by the pPho31 STII variant; a TIR of relative strength 3 was provided by the pPho41 STII variant. Precursor and mature forms of the polypeptide were identified.

In another case, a polypeptide gel was run and stained with Coomassie blue to show secretion of mature NT3 in E. coli under control of variant STII signal sequences. A TIR of relative strength 9 was provided by the pPho31 STII variant; a TIR of relative strength 7 was provided by the pPho21 STII variant; a TIR of relative strength 3 was provided by the pPho41 STII variant; the TIR of relative strength 1 was provided by the pPho51 STII variant. The mature form of the polypeptide was identified.

In still another case, a polypeptide gel was run and stained with Coomassie blue to show secretion of mature RANTES in E. coli under control of variant STII signal sequences. TIRs of relative strength 9 were provided by the pPho31 and the pSTBKPhoA#116 STII variants; a TIR of relative strength 7 was provided by the pPho21 STII variant; a TIR of relative strength 4 was provided by the pSTBKPhoA#81 STII variant; a TIR of relative strength 3 was provided by the pPho41 STII variant; a TIR of relative strength 2 was provided by the pSTBKPhoA#107 STII variant; TIRs of relative strength 1 were provided by the pSTBKPhoA#86 and the pPho51 STII variants. The mature form of the polypeptide was identified.--